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# American Journal Medical Technology

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## A NEW MICRO-PIPETTE FOR BLOOD SEDIMENTATION MEASUREMENTS

By CLYDE BROOKS, M.D.

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A new method for measuring the sedimentation rate of red blood corpuscles, and a new micro-pipette for this purpose, has already been briefly described in a previous publication. Also a brief report has been published giving the results of using this new method in studies of sedimentation in pneumonias. The new micro-technique for blood sedimentation was presented at the Scientific Exhibits, both at the Kansas City meeting of the American Society of Clinical Pathologists, and also at the Fourth Annual Convention of the American Society of Medical Technicians at Excelsior Springs.

The interest manifested in these publications and exhibits indicates a growing appreciation of the value of blood sedimentation tests, as well as an interest in this new micro-technique. So it seems desirable to make further publication in the American Journal of Medical Technology, giving a brief resumé of the principles involved in the method, and giving some points regarding the technique which may be of especial interest to medical technologists.

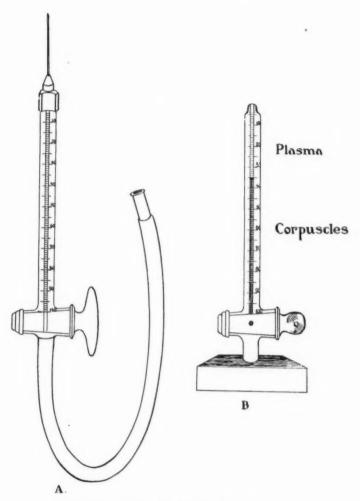
Fig. 1 shows the blood sedimentation pipette: (A) showing the pipette with a hypodermic needle at the end and ready to be filled with blood. (B) shows the pipette inserted in a holder which maintains it in the vertical position, the pipette being filled with blood which is in the process of sedimentation.

The pipette consists essentially of a straight glass tube of small uniform bore which is divided by a stopcock into two limbs. The longer limb is exactly 100 mm. in length and is graduated in millimeters, the zero mark being at the extreme tip, and the 100 mark being exactly at the point where the lumen of the tube is cut by the stopcock. At the end of the graduated limb there is a ground glass tip which fits a Luer hypodermic needle. At the other limb of the pipette a rubber tube is attached. The pipette is used by first attaching the needle to the ground glass tip, and then filling the lumen of the needle and the pipette as far as the 100 mm. mark, or a little beyond the mark, with anticoagulant. Twenty per cent potassium oxalate works very well for this purpose. The anticoagulant which fills the lumen of the pipette is then blown out, leaving a thin film of the anticoagulant solution deposited on the surface of the lumen of the pipette.

In carrying out the method the needle and the pipette are sterilized and dried by drawing air through them, a piece of cotton being placed over the end of the needle to act as a filter. The anticoagulant is kept in a 10 cc. sterile rubber capped vial. With the needle fixed to the ground glass tip, the pipette is filled with anticoagulant by inserting the needle into the vial and drawing the solution up as far as the 100 mm. mark or a little beyond that point. The stopcock is closed and the pipette with needle attached is placed in sterile gauze and carried to the bedside of the patient.

After blowing the contents out of the pipette, using aseptic technique, the needle is inserted into the vein of the patient. The blood by its own pressure fills the pipette very quickly. As soon as the column of blood has reached the 100 mm. mark or just beyond that point, the stopcock is quickly closed. The needle is withdrawn from the vein, and removed from the tip of the pipette. The blood is wiped from the ground glass tip, leaving the meniscus exactly at the tip of the pipette, which is the zero mark.

Thus the pipette is filled with a column of blood precisely 100 mm. in length and 1 mm. to 1½ mm. in diameter, according to the bore of the pipette. The pipette is quickly set up in a holder in the vertical position. If the blood settles very rapidly, readings should be made every minute during the early phase of sedimentation. But if the blood is slow in settling, readings should be made every five minutes. The results are plotted on a chart which is shown in Fig. 2. In order to get complete curves, a two hour period is usually quite sufficient. However, the pipette may be allowed to stand for a twenty-four hour period in order to observe complete sedimentation. However, the readings obtained during the first hour of sedimentation are quite sufficient for most clinical purposes.



Blood sedimentation pipette A.-Ready to be filled B-During sedimentation

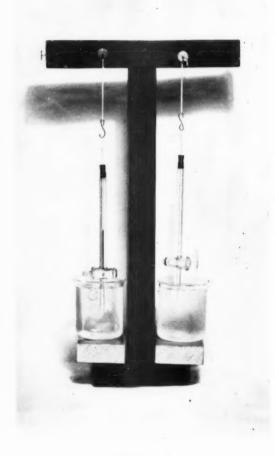


FIGURE 2

Showing pipettes suspended from a stand, and with lower limbs dipping into beaker of water. Pipette at left is in process of sedimentation. Pipette at right is empty.

It is important to have the pipette in as nearly the precisely vertical position as possible, and even a slight variation from the vertical position causes a distinct acceleration of the blood sedimentation. So it is important to have the holders made so that the hole for receiving the pipette is exactly at right angle to the surface of the holder, and it is also equally necessary to have the holder seated on a horizontal plain surface.

Another method which we have used to secure and maintain the vertical position with the pipette is to suspend it by a thread from a suitable stand, as shown in Fig. 2. The method consists of attaching the thread to a short piece of rubber tubing into which the ground glass tip of the pipette can be inserted. This allows the pipette to be suspended by the thread which gives approximately a vertical position. However, drafts cause the pipette to swing. This is obviated by shielding the suspended pipette with wir lbreaks and also by allowing the lower limb of the pipette to extend down into a beaker of water, the water acting as a damper on the movements of the pipette. However, the suspension method is a refinement in technique which is not essential for ordinary clinical purposes.

Another cause of variation in sedimentation rate is the temperature to which the pipette and its contents are exposed during the sedimentation. Cold slows the sedimentation, while warmth accelerates it. Therefore, it is well to carry out the sedimentation tests in a room that is kept at a uniform temperature.

Another important source of error in blood sedimentation is delay in making the observation. The sedimentation rate when taken immediately may be very rapid; but the same blood measured an hour or two later will show a much slower sedimentation rate. After several hours delay the blood may have a normal sedimentation rate. So it is very important to make the sedimentation observation immediately after the blood is drawn from the patient. Of course, in using the method described above there is no delay in starting the observation. The pipette is filled and immediately set up in the vertical position, and the observation made without delay. This is an improvement over other methods where the blood is drawn, measured and mixed with an anticoagulant, carried to the laboratory, and allowed to stand for some time. Then after more or less delay, the blood is thoroughly shaken and put in a graduated tube or cylinder and allowed to sediment. Such results are much too slow, and therefore give erroneous information to the clinician.

Fig. 3 shows various sedimentation curves which may be seen in various infections such as pneumonia. Curve 1 shows a patient who is critically ill with lobar pneumonia. Her chances for recovery are not good.

Curve 2 is from the same patient who is markedly improved.

Curve 3 is a later curve from the same patient who is convalescent.

Curve 4 is a practically normal curve observed in the same patient who has completely recovered from lobar pneumonia.

Curve X is from another patient who is dangerously ill, the prognosis very bad.

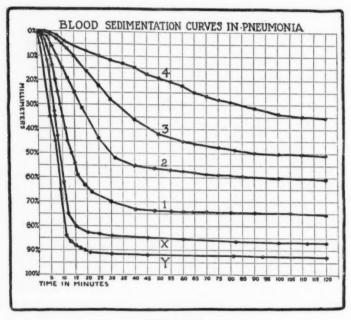


FIGURE 3
Sedimentation Curves in Pneumonia

- Curve 1. Patient critically ill with labor pneumonia.
- Curve 2. Same patient later markedly improved.
- Curve 3. Same patient later convalescent.
- Curve 4. Same patient two months later with normal sedimentation rate.
  - Curve X. Another patient dangerously ill with broncho-pneumonia.
- Curve Y. Same patient three days later. Patient expired a few hours after this test was made.

Curve Y is a later curve from the above patient who is in extremis and very near exitus.

By our micro-method, the total amount of blood used in a single determination is very small: about .075 to .175 cc. Therefore, instead of inserting the needle directly into a vein, the pipette may be filled from a drop of blood made by pricking the finger or the lobe of the ear. This is an advantage on patients who have extremely small veins.

The method is simple, the blood being allowed to flow directly into the pipette, which is immediately set up, and the sedimentation observed. The measurements are automatic: the stopcock closes the lumen of the pipette precisely at the 100 mm. mark, and the removal of the needle from the ground glass tip leaves the meniscus exactly at the zero mark. So the measurements are not only automatic but also are precise.

There is no possibility of leakage nor of air entering into the pipette, because the blood flows into the pipette under its own pressure.

Of course, in order to protect the patient it is not necessary to sterilize the pipette before using it. The use of a sterile needle and sterile anticoagulant would be quite sufficient for this purpose. However, a clean sterile pipette prevents the action of bacteria on the blood. So even after 24 hours or longer, when the sterile blood is emptied from the pipette it is not decomposed. This insures reliable results so far as the sedimentation is concerned and also makes it easier to clean and prepare the pipette for the next observation.

On the whole the method seems to be quite simple in operation. The results are uniformly consistent and reliable. There appears to be a growing interest in blood sedimentation work. It is quite possible that in certain diseases the blood sedimentation test may become as valuable and as frequently used as the blood count.

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## PLAIN FACTS ABOUT BLOOD CULTURES

By ANNETTE M. CALLAN, M.T.

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Before discussing a "successful blood culture," let us define clearly what a blood culture is and what it is for, because every step in the process must be determined by and must lead to the accomplishment of that purpose. Thus far little has appeared in literature beyond directions for technical procedure but, if there be no clear understanding of reasons, long and painstaking labor may result in failure, discouraging at once to the technician and the clinician, the latter especially having received no help toward subsequent procedure.

We all know and appreciate the mental agitation of the clinician, who, when all aspects of medical diagnosis seem obscured, expects the result of a blood culture to solve his difficulty. The surgeon, fearful of the spread of infection, looks to us, hoping that our findings may be negative. Towards both we owe a grave responsibility. We may not lose time nor may we fail in the steps essential for an accurate report. It does not follow that a positive culture is a successful one and a negative culture is an unsuccessful one. The successful blood culture is the positive or negative as it should be; the unsuccessful culture is the direct opposite.

In defining a blood culture, we find therein all things necessary to determine our success or failure. A blood culture is a series of procedures, on the part of a person qualified by training and experience, to collect a sample of blood, under aseptic conditions and by proper technic, and to implant such blood in a suitable medium to an end, that, if a bacterial growth (if present) be obtained, it establishes a diagnosis of septicemia.

Unless qualified by training and experience, collection should not be attempted. We know, however, it is. Collection should not be left to any of several persons,—a physician at his office, a nurse, a technician, an interne, a student et al, and responsibility therefor be placed solely on the bacteriologist. Limit collection and place responsibility for its technic.

Time of collection of the blood is a major factor in the result. It may have to be collected when an ambulatory patient is available. Too often it is a procedure (of necessity) in the early morning, before breakfast, when specimens for chemistry or serology are collected, to save time and to spare the patient. The desirable time for collection is during a chill or at the peak of a fever,—usually in the late afternoon. Countless false negative results must be traced to routine early morning collections,

Precautions for sterility and method therefor and technical manipulation of venous puncture, may be rightly assumed as understood and performed. Many cultures are failures or are improperly evaluated because of some foregoing errors. The pity is that some workers are unconscious of it. Be wary of the staphylococci unless proven, by pigment, haemolysis and enzyme reaction, to be of pathogenic types. Be wary of diphtheroids, likely to be reported as streptococci.

Sample of blood must be reduced to a quantity, not varying from a few drops (all obtainable) to the contents of a syringe, used because obtained. The quantity, for cultures generally, must bear relation to the amount of medium used and the air space of the container. For general work, 1 c.c. of blood to each 25 or 30 c.c. of fluid medium and 1 c.c. of blood to the pour plate is satisfactory. The use of accurately measured 1 c.c. to a plate affords a unit for a colony count, so essential in cases like endocarditis, when aspects of improvement or retrogression are desirable. For such study establish a unit of incubation time,—usually 48 hours. Avoid use of too much blood in the fluid medium. In some cases the antibody content in the serum of a large quantity may cause a serum inhibition to growth. There are special cases where larger quantities of blood are essential to obtain bacterial growth.

Suitable medium varies with the case. For general work we must maintain standard stocks and use them but the aspects of a case being known, these stocks may not be suitable or may have to be varied. All organisms are not recoverable by precisely the same method of collection, in quantity, and implantation on the same media. It is imperative that media to be used be checked by previous incubation to prove its sterility. Until needed for use it should be stored on ice. Optimum pH may be about 7.2 or 7.4. Special organisms require a different range.

The media—both fluid and solid—must be fresh (recently prepared) say within two weeks. There is a great temptation to prepare large stocks and use them until exhausted. They are greatly deteriorated by evaporation and drying. After sterilization, fluid containers should be capped by a circle of paper, fastened down by a single slip-knot of string. This should be used throughout incubation. It prevents dust which might collect on the plug from falling into the container when opened.

The container for the fluid medium is a thing for consideration. The use of test tubes (except for anaerobic work) is not to be recommended as fluid containers. A 250 c.c. round, wide-mouth bottle or 150 c.c. or 200 c.c. Erlenmeyer flask is satisfactory for general work. Such containers should be filled with fluid to 40% or 50% of their capacity. This allows ample air space and adequate depth of fluid even for the streptococci which prefer lessened oxygen. It provides adequate surface for aerobes, a broad bottom surface for growth of lessened oxygen organisms. Observation for latter type of growth is easy by means of a hand glass, otherwise missed because of a clear supernatant broth above sedimented blood line.

The fluid medium (a broth) may be a plain nutrient but an enriched broth is to be desired. The most widely used enrichment is glucose. Recommendations vary from as little as 0.2% up to 1%. The latter amount is likely to be too much for growths of streptococci and pneumococci which are so sensitive to the acidity they themselves form, and while growth is rapid, transfer by subculture to other medium must be prompt or the organisms will die. Amounts of 0.2% or 0.5% are satisfactory for most organisms. Neutrality should be assured by use of 1 gm. of powdered calcium carbonate if long incubation is a necessity before subculture. Bile and ascitic fluid are other forms of enrichment in general use when certain organisms are anticipated.

Plain nutrient agar is used for the blood pour tube. It should be stored in tubes in amounts not less than 10 c.c. or more than 15 c.c. Less than 10 c.c. yields a plate so thin that it dries out during time of incubation. More than 15 c.c. yields a plate too thick for growth of most organisms unless definitely the lessened oxygen type. A thick plate does not afford a proper medium to observe haemolysis or cultural characteristic, such as pigment of the green streptococci or the black of a typhoid colony, by transmitted light.

We may well hail the passage of our early days in the preparation of mussy, time-consuming labor of media preparation. For some, the end product was a total failure. Too much was made, to be used when no longer suitable or to be wastefully discarded when it deteriorated. The prepared media products (such as Difco) are eminently satisfactory. As little as one needs may be successfully prepared. It is granted that certain studies—especially research work, still require the preparation of infusion broths.

The media to use should be at least one fluid and one solid. If time, budget and other factors permit or require it, the fluid and plate in duplicate are certainly desirable as a check on technic and for colony count estimation.

The agar pour plate presents aspects of difficulty especially for the technician who has request to collect a blood culture only occasionally. The c.c. of blood should be accurately discharged by the syringe into the plate. The previously thoroughly melted and then cooled agar from the tube should be added to the dish. By a firm, even, circulating motion, on a flat surface, the plate should be rotated to mix the agar and blood. A perfectly even mixture should result. Some prefer to inject the blood into the tube of melted agar. Then to mix by rolling between the palms. This often results in a frothy mixture and when poured, results first in a collection of air bubbles on the surface which later puncture and cause pin point holes in the plate surface. Such a plate makes a colony count or interpretation difficult or impossible.

Be sure the agar pour tube has been entirely melted, for some time at boiling point. Take to the bedside if necessary, in a glass of almost boiling water. When ready, cool to 48° C. Pour at not less than 45° C. Do not move plate until thoroughly hardened. It should then be turned upside down (and be so incubated) and the moisture rising to the lid may be restored to the medium and not collect on lid and by falling, flood the medium surface.

The method of collecting the blood sample in a few c.c. of sterile sodium citrate is preferred by some, who, after return to the laboratory, inoculate the fluid and solid media. It has some advantages, at times, but the direct inoculation of media by fresh, whole blood is more desirable.

We now approach the important factor of incubation and laboratory manipulation. Incubation at 37.5° C. is usually standard. Do not neglect inspection of temperature of gas and electric incubators each morning and before leaving. Any temperature control may fail, while the small gas-heated type is notorious for variations. Your incubator is more than a mere storage closet. Provide moisture,—a glass of water in the small incubator and one or more large shallow trays of water in the incubator-room type. Cultures may weather a temperature as low as 25° C. if gas goes out over night, but few weather a temperature of 40° C. or more overnight or a week-end.

Incubation time varies with the case. Most common pathogens, suggested or not by the diagnosis, develop promptly and four days

at least are enough to assume that growth is unlikely. Do not be hasty in discarding a negative culture. A week's incubation or even ten days may pick up an unexpected organism which would, otherwise, have been missed. Literature on the streptococci of rheumatic cases recommends incubation for twenty-one days.

If we are doing a fair amount of bacteriology we come to know what to expect for incubation periods of the commoner pathogens; i.e., the streptococcus (haemolytic), growth in 12-24 hours, and haemolysis in 48 hours; the streptococcus (the green), growth in 18-96 hours; the staphylococcus, growth in 12-48 hours; (pigment and haemolysis within 48 hours); typhoid bacillus, growth in about 24-36 hours; miscellaneous organisms of intestinal group—18-48 hours; miscellaneous organisms of respiratory group (B. influenzae, pneumococcus) 24-48 hours; mucosus capsulatus—sometimes very rapid in broth—(12 hours) (average 12-48 hours); meningococcus—(difficult, slow)—72-96 hours.

Growth in both fluid and plate are desirable, as a check on the consistency of type. Growth in broth is likely to be more rapid and luxuriant than in the plate. It may occur in broth only, even in a series of cultures. All studies must be made comparatively on fluid and plate. Turbidity in broth or sign of growth at bottom of fluid must be studied at once. Colonies of like morphology should be looked for in the plate.

Some workers shake their fluid media daily. If this shaking be violent, it breaks the cells and the influence of incubation heat on the released haemoglobin causes a cloudy broth not supported by organisms as a cause. It is desirable to stress the importance of undisturbed fluid media, particularly for growths of streptococci. These grow at the bottom of the medium. If often churned to the surface, into oxygen environment, they may be entirely lost for subculture. The quick, aerobic organisms come up so soon, that restoration of oxygen by shaking is unnecessary. The delicate organisms that need lessened oxygen and are slow growers, do not need this shaking and are harmed by it.

With bacterial growth obtained, interpretation and evalution become the final and most difficult aspects of the study. Experience does much. The less experienced worker (and we may have to add, a careless worker) may throw all Gram positive cocci merely into the staphylococcus group, with an occasional streptococcus for variety. Some are wary of the pneumococci. Unless with a good lead about typhoid, all Gram negative bacilli go into the coli group.

It is desirable to read a culture as readily as we do the headlines of a morning paper. It can be done. From an experience of more than 8 years in handling conservatively 3200 to 3500 blood cultures, the following is offered for direct application in interpretation:

- Study broth carefully, grossly and by glass, daily for evidence of growth; when apparent or suspected;
- Study a hanging drop preparation, do a Gram stain; do a Gram stain from plate colony; read pigment, haemolysis or colony characteristic on plate, and if in accord, make immediate report on the type.
- Make a subculture on a blood plate by streak method; incubate; isolate and carry on to biological identification on appropriate media.

Workers are referred to standard texts for details of cultural characteristics. Merely the major aspects may be presented here.

In broth, the following organisms grow easily, rapidly, with a fairly marked turbidity and darkening of the blood:

Staphylococci, (with a purplish tinge at the bottom if haemolytic), pneumococci (turbid, greenish brown); practically all the negative bacilli (turbid, even throughout):

Streptococci, the viridans of endocarditis,—at first scantily, just above blood sediment, with a clear upper broth;

Mucosus capsulatus-dark, sticky (heat resistant);

Bacillus pyocyaneus-dark, frothy, marked odor in 48 hours;

Bacillus typhosus-light brown, blood layer darkened.

In the plate all the above tend to surface growths rather than deep (but there will be some deep colonies) except the streptococci and the typhoid.

To identify any organism the following must be considered in classifying the organism: motility, spore formation, Gram reaction, gas in glucose, (when indicated), liquefaction of gelatin and coagulation of litmus milk. These studies apply more especially to the bacillus groups. For the micrococcus group (staphylococci, Rosenbach) prove enzymes on Loeffler's blood serum, gelatin and litmus milk. For the pneumococci, cultural and morphological aspects and fermentation of inulin and bile solubility.\* For the Neisseria (Gram negative cocci) the worker is referred to identification by Elser & Huntoon's classification, (available in most texts). For the streptococci, prove by Holman's classification, on lactose, mannite and salacin and the more recently added medium, litmus milk.

<sup>\*</sup>Type against specific pneumococcus serum.

Mention is made of special media, pH, depth of medium and handling methods. For these, the worker is referred to special pH, medium and oxygen requirements for Brucella abortus, in the work of Alice Evans, Huddleson and others, abstracted in modern texts.

For the technic of the massive blood culture (Dr. Herbert Fox, Pepper Laboratory, University of Pennsylvania) essential for obscure streptococcic infection. The method of Cecil for the rheumatic streptococci and of Small, for the streptococcus cardioarthritidis.

The foregoing are difficult for average workers. Much time is required in preparation, handling and manipulation. Contaminations are the likely harvest.

The anaerobic blood culture presents special and difficult aspects. The worker is referred to texts. The results are not eminently satisfying.

The postmortem culture plays a large part in bacteriological procedure in the larger hospitals, especially if connected with teaching institutions. The average postmortem room does not afford a very favorable environment for aseptic collections. Postmortem change and overgrowths of B. coli predominate. The percentage of recovery of primary infective agent is very low—(in pure culture). An analysis of two years' postmortem cultures by the writer shows, in preliminary inspection, many cultures of no value or not indicated, pathological diagnosis considered; a high percentage of coli, with no recovery of the primary infective organism in mixed culture; and only about 5% of recovery of primary organism in pure culture (staphylococci, pneumococci and streptococci) as related to antemortem and postmortem diagnosis.

The organisms most commonly recoverable by blood culture (by no means all of them) are listed as a guide in medical (chiefly acute) and surgical conditions.

Furuncles, cellulitis, etc.: staphylococci, streptococci, B. proteus; B. pyocyaneus; B. coli.

Osteomyelitis: Staphylococci, streptococci, B. pyocyaneus, B. proteus.

Brain abscesses, meninges: Streptococci, pneumococci, staphylococci, meningococci, B. pyocyaneus, B. coli and B. typhii (occasionally).

Chest and respiratory tract: Pneumococci, streptococci, staphylococci, B. Friedländer, B. pyocyaneus (abcesses).

Nose (and sinuses): Staphylococci, streptococci, pneumococci, meningococci, B. Friedländer, B. influenzae, B. pyocyaneus, B. coli.

Throat: Comparable to above.

Mastoid and ear: Streptococci, pneumococci, staphylococci, B. proteus, B. pyocyaneus, B. influenzae, B. Friedländer.

Intestinal: B. typhi, B. coli, B. pyocyaneus, B. proteus and (more rarely), B. dysenteriae, Br. abortus, streptococci (abdominal surgery).

Genito-urinary: B. proteus, B. pyocyaneus, B. coli, staphylococci, streptococci.

To conclude: All aspects of manipulation, media, cultivation, environment, gross inspection, microscopy and biological proofs are so closely related and interdependent, that no study can be complete and successful unless utmost care and thoughtful judgment be applied to every case,

## HEMOLOGIC OBSERVATIONS ON THE ANEMIAS AND LEUKEMIAS\*

## II. RETICULOCYTE RESPONSE IN PERNICIOUS ANEMIA

By E. A. SHARP, M.D. and E. M. SCHLEICHER, A.B.

In the initial report (1) of this series the myeloid reaction during relapse and induced remission of pernicious anemia was discussed. The second interesting hemopoietic phenomenon incident to recovery of the blood in Addisonian anemia is the occurrence of reticulocytosis.

Nature and Morphology of the Reticulocyte—The reticulated red blood cell is an immature erythrocyte normally present in the blood stream in a percentage concentration ranging from 0.8 to 2.5. The reticulum consists of granulo-filamentous structures having an affinity for vital stains. The precise nature of the mitochondria in the reticulocyte is unknown, but it is readily destroyed by fixatives containing ether, alcohol, chloroform, et cetera. The affinity of reticulum for vital stains favors a protoplasmic rather than a nuclear origin of this cellular structure.

In his early study of reticulocytes Hawes (2) found that they are frequently larger than non-reticulated erythrocytes in the same preparations. Persons (3) has reviewed this subject fully. He showed that reticulocytes in normal blood and in the blood of pernicious and non-pernicious anemia have a mean diameter greater than that of the normal erythrocyte. Wintrobe (4) has reported observations suggestive of increased corpuscular volume resulting from the presence of an abnormal number of reticulocytes.

Buckman and MacNaugher (6) studied fragility of reticulocytes found in pernicious anemia and concluded that their fragility is slightly decreased. In our experience the fragility of composite specimens of red blood cells during severe relapse of Addison's anemia varies with the patient; hence, this property of the reticulocyte would be expected to vary individually.

<sup>\*</sup>From the Anemia Laboratory, Out-Patient Department, Harper Hospital, Detroit, Michigan.

The life cycle of the reticulocyte has been studied extensively since greater attention has been focused on this cell as a result of modern study of pernicious anemia. Whether it matures in the peripheral circulation remains controversial. In view of this uncertainty a discussion will be reserved for the future.

Classification of Reticulocytes—Engel (5) has classified reticulocytes from a morphologic standpoint. He concluded that four distinct classes are detectable. Summarized briefly Engel's four groups are as follows: Class 1, containing dense reticulum occupying one-third to one-half of the area of the cell are very immature; Class 2, showing one-fourth or one-fifth of the cell area are immature; Class 3, when showing about the same amount of network as in Class 2 but in a loose, branched or disorganized arrangement are maturing cells; and Class 4, show a majority of reticulum as marginally placed remnants, with a fine thread like process in the cell body occasionally. This reticulocyte is a mature form.

While Engel's classification is the classical, in our blood studies we regard as Type A Engel's Classes 1 and 2 since their mean diameter and concentration of reticulum are similar. In our experience the mean diameter of Engel's Classes 1 and 2 is usually between 11 and 12 microns as measured by a Leitz Echelon ocular micrometer. Since Engel's Classes 3 and 4 manifest a decreased mean diameter, usually about 9.0 microns, and a reduced amount of reticular structure we are accustomed to regard this group as Type B.

Reticulocyte Response Induced by Therapy-With the introduction of liver by Minot and Murphy for the treatment of pernicious anemia they postulated that an increase of reticulocytes during remission induced by liver was in inverse proportion to the concentration of the erythrocytes at the pretreatment stage. Stated in another way, the greater the reduction in red blood cells the higher the concentration of the reticulated erythrocytes during early induced remission.

Minot and his associates (7), therefore, prepared a mathematic formula for calculating the maximum rise of reticulocytes for various pretreatment red blood cell levels following adequate treat-These numerical determinations are usually referred to as "calculated maximum reticulocyte percentages." The original values have been modified to conform to observations made on the treatment of pernicious anemia with stomach tissue and, also, for remission induced by parenteral liver extract. While the "calculated maximum reticulocyte percentages" vary slightly with the form of antipernicious anemia therapy, the mode of a response,

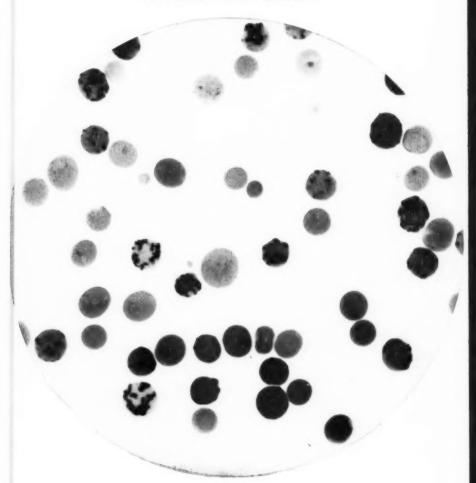
when charted graphically, is substantially the same when optimal amounts of any potent antipernicious anemia preparation is given (Figure 2). The maximum concentration of reticulocytes usually appears about the sixth or seventh day of treatment. The following day there is an appreciable decrease in the number of reticulocytes while on the third post-critical day a secondary rise may occur giving a "notched" mode to the descending limb of the numerical graph. The time elapsing from the beginning of the reticulocytosis until the return to a normal percentage varies considerably but ranges in most cases from twelve to sixteen days.

Figure 1 represents the blood picture at the height of reticulocyte production, which occurred on the sixth day of treatment. It will be seen that all types of reticulocytes appear during the crisis. Some of the immature cells show wreath forms of reticulation, others completely filled with the granulo-filamentous threads while a few are more mature as judged by the marginally placed reticular The appearance of a few reticulocytes containing dense reticular structure (Type A) in any blood picture is a presumptive criterion of increased hemopoietic activity. Conversely, when all the immature erythrocytes in an individual picture continue to show only remnants of reticulum (Type B) reduced activity in the blood forming system can be assumed. Frequently, as early as the second day of treatment of severely relapsed cases of pernicious anemia densly reticulated erythrocytes will appear in the picture. This observation presages remission of the anemia, hence is of importance in the daily record of blood findings when the therapy or the patient's responsiveness is in question.

Of interest at this point also is that the morphology of the non-reticulated erythrocytes during the stage of reticulocytosis may not have changed materially when compared with the pretreatment picture. Megaloblastosis, macrocytosis, bizzare forms, polychromatophilia, poikilocytosis, anisocytosis, et cetera, may persist until the phase of reticulocytosis has supervened. Ordinarily, erythrocytic maturation begins when the stage of reticulocyte production is declining; the total red blood cell count minus the absolute concentration of reticulocytes will show an absolute increase of mature erythrocytes.

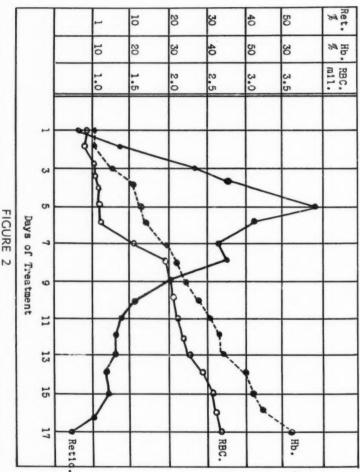
The total blood count seldom increases appreciably, however, in absolute numbers of erythrocytes during the early phase of reticulocyte production even though the absolute number of reticulocytes is greater. Occasionally the total red blood cell count decreases during the initial stage of reticulocytosis, which is probably due to abnormal consumption or retarded maturation of primitive erythrocytes. It would appear, then, that the erythrogenic function of the

## HEMOGRAM IN COLOR



## FIGURE 1

Maximal reticulocyte response during induced remission of severely relapsed pernicious anemia. The reticulocytes in the hemogram show varying amounts of reticulum, the denser concentration being in the most immature cells. While not invariable, the younger reticulocytes are larger than the older forms. The photograph was taken on the sixth day of treatment when the concentration of reticulated red blood cells was 35 per cent.



Hemograph showing a reticulocyte response after adequate treatment of a case of severely relapsed pernicious anemia. The pretreatment red blood cell level was about 900,000 per cu. mm. The actual reticulocyte response was 58 per cent, which occurred on the fourth day of treatment. An early reticulocyte crisis and an excessive rise are not uncommon; hence, daily hemologic observacalculated maximum reticulocyte percentage for this red blood cell level is about 38 per cent. tions should be made in order to avoid missing the height of reticulocytosis,

hemopoietic system in early induced remission is principally devoted to producing immature erythrocytes, hence maturation does not follow until a substantial number of primitive red blood cells have been developed.

General Considerations—It is of interest to record that the bone marrow pattern at the time of the reticulocyte crisis may continue to show myeloid cells of the hypermature type described in our previous communication (1).

Bone marrow removed from cases of pernicious anemia on the day the reticulocytes attained a maximum level shows multi-lobed neutrophils together with stab and juvenile forms. It will be recalled, also, that the left myeloid shift ensues about the same time that the maximal reticulocyte response is observed in the blood pattern. There is, therefore, concomitant hyperneocytosis exhibited by both the red and white blood forming structures as the result of adequate treatment of pernicious anemia.

In all biologic phenomena there are variables to be considered. This fact cannot be excepted in disposing of the subject of erythropoiesis in Addison's anemia since the classical reticulocyte response to adequate treatment of severely relapsed anemia as described above may not ensue. One of the common aberrations is when the maximum concentration of the immature red blood cells at times fails to reach the "calculated" level. Such a deficient quantitative response may be due to numerous clinical conditions, but the technic of making the reticulocyte determinations also should be scrutinized. Solutions of brilliant cresyl blue commonly used for staining the reticulum, deteriorate rapidly due to deaminization; hence, many cells may remain unstained. Fresh stain and repeated counts should be made as a check on the causes of low counts. It may be advisable to make reticulocyte counts every two hours during the day as the time for the expected reticulocyte crisis approaches, in order to determine the erythropoietic trend. It is, of course, obvious that technic may be correct in every particular on all occasions, yet the critical rise is missed because only one count is made in 24 hours.

## Summary

The quantitative reticulocyte response in early remission is a valuable hemologic criterion for predicting other hemopoietic changes in the blood. Subsequent to this phenomenon the mean diameter of the red blood cells tends toward the normal, megaloblastosis disappears and hemoglobin synthesis ensues. Careful consideration should be given to all these phenomena each day

during the initial treatment period of pernicious anemia not only in order to assess the accuracy of the salient technical procedures employed but also to demonstrate the value of constant hemologic observation to the clinician.

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## A COMPARISON OF THE KOLMER-WASSER-MAN, KAHN AND JOHNS PRECIPITATION TESTS ON BLOOD SEROLOGY

By HERMINE TATE, M.T. and E. M. ROBARDS, M.D.

From the East Louisiana State Hospital, Jackson, La.

As we step back a few years we learn that many physicians practiced medicine without the aid of a laboratory. The complexity of the various tests, as well as the need of elaborate and expensive equipment, put them beyond the reach of the average physician. The Doctor usually formed his diagnosis according to clinical and physical findings.

Medical science is perfecting itself more and more as time passes on. Many of the laborious laboratory techniques have been greatly modified. Today we have modern equipped laboratories with pathologists and competent technicians within the reach of practically every physician, while some physicians have simple equipment in their offices and do some of their own minor tests.

The history of syphilis in general begins in controversy—whether the Indians found on the American continent were the first to be infected with the disease; or whether the Europeans brought it over to America. At any rate it did not receive widespread attention until after 1492.

As late as the latter part of the 18th Century scientists were working on a method of diagnosing syphilis by the examination of the blood. Bordet and Gengou's first application of the complement fixation which was carried out with a suspension of B. pestis was in 1898. It is interesting to note that Bordet and Gengou, according to Metschnikoff, were the first to see the Spirochaeta pallida. But in the race for the development of the possibilities of this new type of diagnostic procedure they again lost the preeminence that would have come from the initial application of the complement fixation test to the diagnosis of syphilis. Instead, the distinction fell to Wassermann, Neisser, and Bruck.

In 1907 Wassermann published his first literature and technique on the complement binding reaction. Whole lifetimes of labor were bestowed upon the perfection of this test, resulting in numerous modifications of the original Wassermann reaction. Progress in diagnostic procedure was rather slow at first. But within the past decade or more there has been a marked advance in the serological diagnosis of syphilis. Partly responsible for this is the increasingly widespread use of various precipitation tests—such as the Kahn, Johns, Eagle, Kline, Hinton and others; there being a sharp contrast in the simplicity of these as compared to the time-honored complement fixation reaction. As for the comparative value it is a field which has brought and is bringing forth much discussion.

In the complement fixation method some laboratories practice the use of more than one antigen, these varying in sensitivity. In my estimation this does not adequately check against a false positive or negative reaction, whereas the use of more than one method will tend to lessen a false diagnosis of syphilis. There are few laboratory procedures whose inevitable margin of error cause more social and personal suffering than that of serology.

When patients are admitted to the East Louisiana State Hospital we do a Kolmer-Wassermann, Kahn, and Johns precipitation test for the detection of syphilis in the blood. Complete records for the past three years show that 20.4 per cent of the general admissions have the said disease. Of these infected individuals 40.3 per cent have constitutional syphilis, while 58.7 per cent are cerebrospinal. 8 per cent fluids not done due to physical condition of patient. 35.3 per cent of our general admissions over this period of time are colored patients, syphilis being much more prevalent in that race. Also the proportion we find in the per cent of cerebrospinal syphilis, as compared to constitutional syphilis, is quite different from that which one would find in a series of individuals other than in a mental hospital:

## Brief Techniques

From 4 to 5 c.c.'s of blood is aseptically drawn from vein of the patient's arm, and examined by the three serological methods within 24 hours after it is obtained. All glassware used is chemically clean, dry, and sterile. Bacterial contamination of the blood, either through carelessness in drawing a specimen, or through delay in testing, is occasionally responsible for a false positive or anti-complementary result.

#### 1-Kolmer-Wassermann:

a-Kolmer cholestrolized antigen.

b-Anti-sheep ambroceptor.

c—Complement (consisting of the fresh pooled serum from not less than three guinea pigs).

d-Defibronated sheep cells.

Twelve to eighteen hour ice-box fixation method.

### 2-Kahn Precipitation:

Into three test tubes place .05; .025; and .0125 c.c.'s, respectively, of properly diluted Kahn antigen. Add to each of the three tubes .15 c.c.'s of inactivated serum. Shake tests vigorously for three minutes. Add .5 c.c.'s of physiological salt solution; shake for few seconds and read.

#### 3-Johns Precipitation:

Smear .15 c.c. of clear un-inactivated serum on middle one-third of a clean greaseless microscopic slide—let thoroughly dry at room temperature which takes several hours. Drying can be greatly hastened by use of a hot water bath. To dried serum add .2 c.c. of properly diluted Johns antigen. With the aid of a toothpick spread antigen over serum, and by a circular rubbing motion dissolve all dried particles of serum with the antigen. Let stand 5 minutes and read.

In the following series of tables I will endeavor to show you the effect of which various amounts of anti-luetic therapy have upon the three serological tests used.

#### TABLE I

This consists of the routine blood examinations on 2,159 patients, by the Kolmer-Wassermann, Kahn, and Johns precipitation methods for sero-diagnostic purposes. A large per cent of these individuals are non-syphilitic. Some are known to be infected with this disease, having been under anti-luetic treatment prior to present admission. Others have syphilis at various stages giving a history of no anti-luetic therapy. Seven of the eleven individuals giving a anti-complementary Wassermann reaction have proved to be syphilitic; the remaining four were evidently caused by error in technique. Also in this series we found six patients who were by clinical, neurological, as well as by spinal fluid examinations, diagnosed paresis, giving a repeated negative blood test by all three methods. Unfortunately I do not have the history of these individuals as to the amount of treatment given.

Table I
Bloods from 2,159 patients on general admission.

	Kolmer Wass. Pct.	Kahn Pct.	Johns Pct.	
Negative	79.7 %	79.2 %	79.0 %	
Doubtful	0.27%	0.74%	.88%	
Positive	19.4 %	18.4 %	18.7 %	
Anti-comp.	.5 %	******	******	
Not done	******	1.4 %	1.3 %	

#### TABLE II

This group consists of 110 individuals being infected with syphilis, either constitutional or cerebrospinal, who have had from 2 to 8 weeks of anti-luetic therapy. Some of these patients were picked up as we tried to use them as a positive control after anti-luetic treatment had been started; others gave a definite history as to the number of treatments prior to admission; while a few others were checked special for this study of comparisons. This presents the effect that the first few anti-luetic treatments have upon the three serological tests.

Table II

Bloods from 110 constitutional and cerebrospinal syphilitic patients after 2 to 8 weeks of anti-luetic treatment.

Ko	lmer Wass.	Kahn	Johns
	Pct.	Pct.	Pct.
Negative	2.7%	15.4%	22.7%
Doubtful	2.7%	16.3%	19.6%
Positive	90.9%	65.4%	55.6%
Anto-comp	3.6%	********	********
Not done		2.7%	1.8%

#### TABLE III

Here we are dealing with the type of infection with which the general practitioner is more familiar. The incarceration of these patients in a mental hospital is not due to syphilis, although in some cases it does tend to aggravate the psychosis. This series consists of a group of bloods from 130 individuals with constitutional syphilis, who have had from 18 months to 4 years of anti-luetic therapy. Spinal fluid examinations made from time to time continue to be negative. Blood serology is done every 6 months for the prognosis of the disease.

Table III

Bloods from 130 constitutional syphilitic patients after from 18 months to 4 years of anti-luetic treatment.

K	olmer Wass. Pct.	Kahn Pct.	Johns Pct.
Negative	63.0%	43.8 %	40.0%
Doubtful	18.4%	23.0%	26.1%
Positive	15.4%	27.6%	30.0%
Anti-comp.	3.0%	*****	******
Not done	******	5.3 %	3.8%

#### TABLE IV

Now we come to the syphilitic who usually falls into the hands of the neuro-psychiatrist for treatment, as individuals so infected are generally found in the psychopathic wards, or hospitals for mental diseases. This group consists of 160 individuals with cerebrospinal syphilis, having had hospitalization and treatment for the past 18 months to 4 years.

#### Table IV

Bloods from 160 cerebrospinal syphilitic patients after 18 months to 4 years of anti-luetic treatment.

Ko	olmer Wass. Pct.	Kahn Pct.	Johns Pct.
Negative	57.5%	26.8%	21.2%
Doubtful	5.1%	17.9%	22.5%
Positive	36.2%	53.7%	54.3%
Anti-comp	1.0%	1.2%	1.8%

#### Results

In our first series of results (which consisted of general admissions) we found that the Kolmer-Wassermann, Kahn, and John precipitation tests checked quite favorably. The majority of the discrepancies in that table of blood examination were found to be from syphilitic individuals. These later gave a history of having had anti-luetic treatment prior to admission to hospital, when the serological tests were made.

The second series consisted of bloods from known syphilitic individuals, who had just received from 2 to 8 weeks of anti-luetic therapy. From these we obtained a negative or doubtful precipitation test (more pronounced in the Johns than in the Kahn) without any great change in the Kolmer-Wassermann reaction. I did not segregate these individuals as to the stage of infection.

From our series of constitutional syphilitic individuals, having received extensive anti-luetic therapy, we found by far more negative reactions from the Kolmer-Wassermann method than by either of the precipitation tests.

In our series of blood examinations from treated cases of cerebrospinal syphilis, we obtained practically the same ratio of comparisons as we did from the treated constitutional syphilitic individual. Except, one found more of that which is known as Wassermann fast bloods. They stay positive year in and year out regardless of the type of frequency of anti-luetic therapy given.

Therefore, in order to get a cured or an arrested case of syphilis, diagnosed by our serological laboratory, the clinician looks for a negative report from the precipitation tests as well as from the Wassermann reaction. I consider the Johns to be a simple and economical precipitation test agreeing quite favorably with the Kahn. But with all the popularity of the numerous precipitation tests, the Wassermann reaction probably remains the one method most widely used for sero-diagnostic purposes.

### Summary and Conclusion

1—Brief techniques are given on the Kolmer-Wassermann, Kahn. and Johns precipitation methods.

2—Serological examinations tabulated on 2,159 individuals. This included all patients as they were admitted to the East Louisiana State Hospital over a period of three years.

3-Blood serology was tabulated, on 110 syphilitic individuals, by the Kolmer-Wassermann, Kahn, and Johns precipitation tests. These patients had received from 2 to 8 weeks of anti-luetic therapy.

4-A study of the comparisons of the Kolmer-Wassermann, Kahn, and Johns precipitation tests were made on bloods from 130 constitutional syphilitic individuals, after extensive treatment had been given.

5—Tabulations and comparisons of the Kolmer-Wassermann, Kahn, and Johns precipitation tests were made on 160 cerebrospinal syphilitic individuals after extensive anti-luetic treatment.

In view of the above statistics, my conclusion is that the first few anti-luetic treatments given to a syphilitic individual cause a negative or doubtful reaction quicker by the two precipitation methods than by the Kolmer-Wassermann reaction. But as the course of treatment is continued the precipitation tests return positive, and under rigid treatment stay positive longer than the Kolmer-Wassermann reaction. And that no syphologist or laboratorian should rely on a single test for the diagnosis or prognosis of syphilis.

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## **EDITORIAL**

### VALUE OF AFFILIATION

One very reasonable question any registered Medical Technologist must surely ask himself or herself some time is, "Why should I become a member of the American Society of Medical Technologists and what advantage will it be to me?"

First of all the Society is the only national organization conducted entirely within the code of ethics as outlined by the American Society of Clinical Pathologists. Anyone who has chosen laboratory work as a permanent vocation and is able to qualify and becomes a registered Medical Technologist after passing the required examination of the Board of Registry of the American Society of Clinical Pathologists, realizes that he now has a definite professional rating.

Second, the registered Medical Technologists in forming a Society, have an organization purely for individual advantage, the benefit derived therefrom being proportional to the activity of each member. The chief reason for organizing being to sponsor, to encourage and to help every conscientious Medical Technologist to use their initiative, to keep well informed of the latest procedures and to help solve those difficult problems that arise from time to time.

Third, the official organ of the Society, "The American Journal of Medical Technology," extends a medium of publication to all those who wish to publish material of value. The Society urges its members to contribute to the Journal those articles which are of real value, as many phases of laboratory work are represented by members of the organization.

Briefly, one may say, for a laboratory worker to realize the utmost from his chosen vocation he must enter into co-operation with those of his own profession. The American Society of Medical Technologists offers this opportunity through its membership and Journal. Every year just after the annual meeting of the American Society of Clinical Pathologists and during the annual session of the American Medical Association, the Society convenes formally to hear outstanding scientific papers and to transact the business of administration. Members are urged to attend this meeting.

Although organized only a few years, the Society is rapidly gaining recognition through the Journal and its members, which in turn results in an ever-increasing privilege of fellowship.

## **NEWS AND ANNOUNCEMENTS**

## REGISTRY OF MEDICAL TECHNOLOGISTS OF THE

#### AMERICAN SOCIETY OF CLINICAL PATHOLOGISTS

Thirty-five hundred registrants received the fifth edition of the annual Roster of the certificate holders of the Registry in December. We are confident that all registered M.T.'s will take a just pride in having their names appear in this edition.

The fall examinations were conducted simultaneously throughout the United States, Canada, Hawaii, and the Canal Zone on October 29. The examining Board used the following questions in the written test:

### I. Urinalysis

- Describe (a) a recognized quantitative test for the determination of urinary sugar. (b) a recognized quantitative test for the determination of urinary albumin.
- Describe five (5) pathological elements which may be found in a microscopic examination of urine.

## II. Clinical Bacteriology

- How would you differentiate between (a) B. Coli; (b) B. Typhosus; (c) B. Paratyphosus A; and (d) B. Paratyphosus B?
- 2. (a) What are the cultural characteristics of the diphtheria bacillus? (b) What morphologic and staining characteristics are of importance in their identification?

## III. Hematology

- (a) Give a procedure for the counting of blood platelets.
   (b) What is meant by "clot retraction time" and how is it determined?
- Suppose you are in a home and have forgotten your white cell pipette, state how you could use the red cell pipette to do a leukocyte count and give your computation in detail.

### IV. Serology

 Give the details involved in conducting pretransfusion tests for: (a) Determination of the blood group. (b) Cross agglutination for determination of compatibility. (Use international — Landsteiner — nomenclature either alone or in addition to any one of the two numerical nomenclatures.)

Name the controls that are used in the complement fixation test for syphilis (Wassermann test). State the purpose of each control.

V. Blood Chemistry

1. Name the non protein nitrogen constituents of the blood.

VI. Tissue Technique

1. Name two standard staining methods for tissues.

#### NATIONAL

On October 2, 1936, Sarah McCarty sailed for Edinborough, Scotland, to complete her medical training, thereby resigning her post as Chairman of the Program Committee. We shall miss her loyal support and enthusiastic work in the Society and extend our best wishes for success.

Luella Gifford, who has consented to "carry on," as program chairman, is now busily engaged in the work necessary to give us a worth-while meeting next June.

Program Committee, 1936-37:

1. Miss Luella Gifford, 339 Boush Street, Norfolk, Va.

- Miss Annette Callan, 316 Carver Hali, Frankford, Philadelphia, Pa.
- Miss M. Hermine Tate, E. Louisiana State Hospital, Jackson, La.
   Miss Frances Chamberlin, 2315 S. 17th St., Lincoln, Nebraska.
- Miss Anna Mary H. Falck, Lancaster General Hospital, Lancaster, Pa.

It was with regret that we were obliged to accept the resignation of Sister M. Joan of Arc from all offices and active participation in the American Society of Medical Technologists. Sister M. Joan tendered her resignation on October 13, 1936, being forced to do so because of ill health. Sister Joan was President-Elect, an ex-officio member of both the Constitution and Affiliation Committees. We hope for a speedy recovery and that we may again have her able assistance in the future.

#### STATE

#### Illinois

The fall meeting of the Illinois Society of Clinical Laboratory Technicians was held in Springfield, October 2-3, 1936. The following program was presented:

#### FRIDAY, OCTOBER 2—Evening Session Auditorium, Centennial Building

Presiding, Mary A. Kennedy, Veteran's Hospital, Danville

8:00 President's Greeting—Margaret J. Burgess, Mercy Hospital, Urbana.

8:10 Methods for Isolation of Typhoid Bacilli from Fecal and Blood Specimens, H. J. Shaughnessy, Chief, Division of Laboratories, State Department of Public Health, Springfield.

8:30 Methods of Propagating Filtrable Virus, C. A. Brandly, Laboratory of Animal Pathology and Hygiene, University of Illinois, Urbana.

8:50 Some Common Allergies, F. M. Clark, Department of Bacteriology, University of Illinois, Urbana.

SATURDAY, OCTOBER 3—Morning Session Presiding, James Yeager, Waukegan

9:00 Business Meeting.

9:30 Report of National Convention at Kansas City—Christine C. Seguin, Niles Center.

10:00 Recent Contributions to the Bacteriology of the Diphtheria
Bacillus with Special Reference to the Incidence and
Clinical Significance of the Various Types in Illinois—
T. C. Grubb, Division of Laboratories, State Dept. of
Public Health, Springfield.

10:30 Proposed Revisions in Incubation Temperatures in Milk Analysis—S. V. Layson, Division of Sanitary Engineering, State Dept. of Public Health, Springfield.

11:00 Water and Sewage Bacteriology and Microscopy — E. S. Clark, Division of Sanitary Engineering, Department of Public Health, Springfield.

Afternoon Session

St. John's Hospital—Eighth and Mason Streets Presiding, Dorothy Shaw, Methodist Hospital, Peoria

1:30 Demonstrations:

Methods of Determining Blood Sugars—F. W. Light, M.D., Pathologist, St. John's Hospital, Springfield.

Blood Platelet Counts—Sister Willibalda Dasenbrock, St. John's Hospital, Springfield.

A Shortened Method for Matching Blood Donors for Transfusions — Fanny Warnock, Burnham City Hospital,

Champaign.

Blood Cyanates — Ruth Chapin, Carle Hospital Clinic,
Urbana.

Evening Session
Elks' Club, 509 South Sixth Street





6:00 Dinner.

7:30 Round Table Discussion and Demonstration

Presiding, F. Warnock, Burnham City Hospital, Champaign.
Officers of the Society are: President, Margaret J. Burgess, M.T.,
Mercy Hospital, Urbana; Vice-President, Alma E. Tillotson, M.T.,
Municipal T. B. Sanatarium, Chicago; Secretary-Treasurer, Myrtle
Sand, M.T., Cook County Hospital, Chicago.

Ohio

Alice Finnin, M.T., President of the Ohio Society of Medical Technologists, and Martha Andes, M.T., secretary, attended a business meeting of the Ohio Hospital Association Tuesday afternoon, at the Deshler Wallick Hotel, Columbus.

Plans were made to hold the fourth annual State Convention of Medical Technologists in conjunction with the Ohio Hospital Association, Wednesday, April 15, 1937, at the Deshler Wallick Hotel,

Columbus.

#### H'isconsin

On Saturday, October 10, 1936, thirty registered Medical Technologists, engaged in Laboratory work in Wisconsin, met in Madison and organized the Wisconsin Association of Medical Tech-

nologists.

The meeting was called to order by Lona Jacobson, M.T., of LaCrosse, with Laura Bates, M.T., of Madison, acting as secretary. After the invocation by the Rev. L. B. Mosely, of the First Baptist Church, Madison, Grace T. Crafts, R.N., Superintendent of the Madison General Hospital, welcomed the group into the ranks of the organized medical societies of the state, and assured us of the hearty good wishes and co-operation of the State Nurses' Association, and the Tri-State Hospital Association, of which she is a vice-president.

Dr. W. D. Stovall, Director of the State Laboratory of Hygiene,

was the guest speaker of the morning.

The afternoon session, devoted to the business of the day, saw the constitution adopted and the officers for the year 1937 elected: President, Laura Bates, M.T., Madison; President-elect, Lona Jacobson, M.T., LaCrosse; Vice-President, Sr. M. Ephram, M.T., Manitowoc; Secretary, Isabelle Gallagher, M.T., Stevens Point; Treasurer, Catherine Barron, M.T., Milwaukee; Historian, Bernice Gage, M.T., Madison; Sargent-at-arms, Stephan Weber, M.T., Milwaukee.

Directors to assist the first five officers representing the four districts of the state: District 1—Sr. M. Bernadette, M.T., Janesville; District 2—Sr. M. Venantia, M.T., Appleton; District 3—Sr. M. Corona, M.T., LaCrosse; District 4—Ruth Mentges, M.T.,

Wausau.

An informal dinner was held in the evening.

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